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# Batch, fed-batch and CSTR reactors as cultivation systems to acclimate ammonia tolerant methanogenic consortia

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## Introduction

Ammonia-rich substrates are known to inhibit anaerobic digestion process and it is estimated that many full-scale biogas reactors are seriously affected by ammonia toxicity leaving up to 1/3 of their methane potential unutilized. Bioaugmentation of ammonia tolerant methanogenic consortia has been proposed as a promising method to attack this challenge.

A major bottleneck for a successful bioaugmentation of ammonia inhibited systems is the accessibility to ammonia-tolerant methanogenic cultures. To date, no study can be found indicating the most efficient method (in terms of incubation time, TAN and FAN levels achieved) to acclimatise ammonia tolerant methanogenic consortia.

## Aim

The aim of the present study was to assess the efficiency of the three different cultivation methods (i.e. batch, fed-batch and CSTR) to acclimatise methanogens to high ammonia levels. In batch experiment, both stepwise and direct-exposure were tested.

## Materials & Methods

Three different experiments (i.e. batch, fed-batch and CSTR) were performed to compare the efficiency of the different cultivation modes, on acclimating two initial inocula to high ammonia levels. In all three experiments, the inocula were incubated at its original TAN levels for lab-scale environment adaptation and determination of the uninhibited methane production.

## Materials & methods

**Batch experiment.** Two tests were performed during batch experiment: 1) direct-exposure of the uninhibited inoculum to different ammonia levels 2) stepwise exposure to increasing ammonia levels through successive batch cultivations.



**Fed-batch experiment.** Two fed-batch reactors were used, one for the mesophilic and one for the thermophilic inoculum. Starting with 45 mL inoculum, an exponential feeding strategy was performed with an OLR of 0.5 g HAc L<sup>-1</sup> d<sup>-1</sup>.

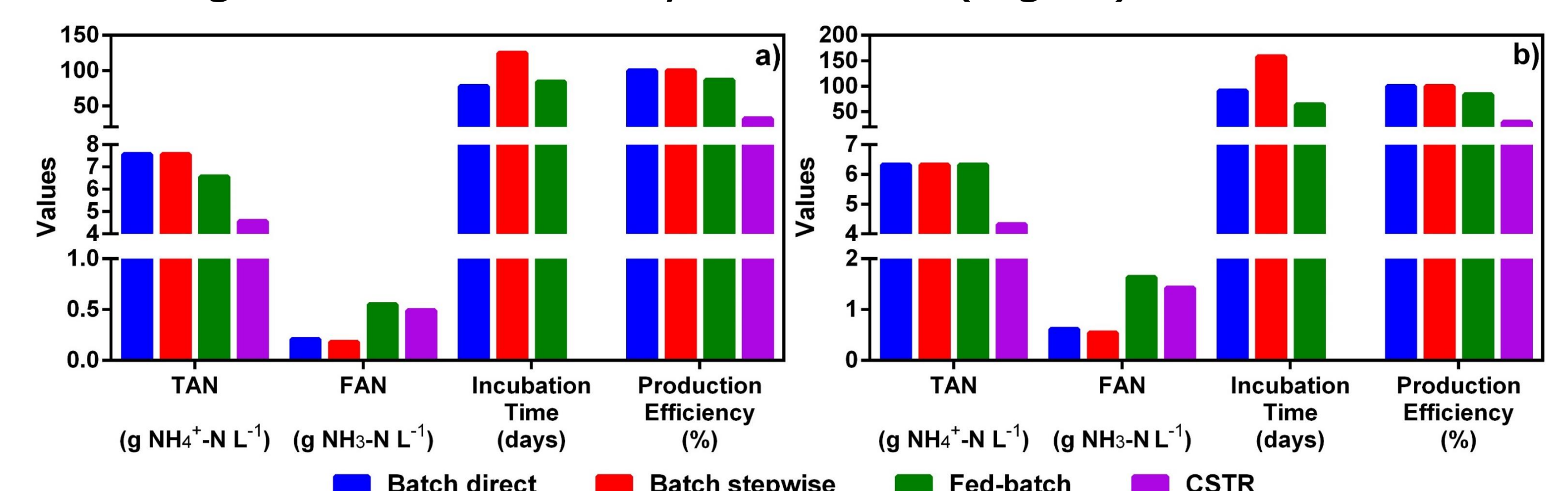


**CSTR experiment.** One mesophilic and one thermophilic CSTR reactors were used with 20 and 15 days HRT, respectively. Each reactor had 1.8 L working volume and an OLR of 0.5 g HAc L<sup>-1</sup> d<sup>-1</sup>.



## Results

The methane production in all **batch and fed-batch reactors reached the theoretical methane production contrary to both CSTR reactors that did not pass the second acclimation step** probably due to wash out of key microorganism inhibited by ammonia (Fig. 1).



**Fig. 1.** Comprehensive comparison between the three different acclimation methods for a) mesophilic and b) thermophilic inocula.

## Results

The fed-batch had the shortest incubation period compared to batch reactors at the highest FAN levels for both mesophilic and thermophilic conditions. Even though mesophilic batch direct-exposure method had a few days shorter incubation time than fed-batch, the FAN levels of the mesophilic fed-batch reactor was 164% and 138% higher than batch direct-exposure method, respectively. The thermophilic fed-batch acclimation method had more than 40% shorter incubation time compared to batch acclimation.

Concerning the production efficiency, only 30% of the theoretical production was detected during CSTR acclimation method, while more than 83% was achieved in batch and fed-batch reactors.

## Conclusions

- Fed-batch is the best ammonia acclimation method based on production efficiency, incubation time and TAN levels.
- CSTR reactors are not suitable systems to efficiently acclimatise ammonia tolerant methanogenic consortia.
- Fed-batch is an efficient method to cultivate ammonia-tolerant methanogens that can be bioaugmented in biogas reactors to alleviate ammonia toxicity effect.

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